J. Glob. Innov. Agric. Soc. Sci., 2015, 3(2-3): 68-71.
ISSN (Online): 2311-3839; ISSN (Print): 2312-5225
DOI: 10.17957/JGIASS/3.2-3.717
http://www.jgiass.com

LARVICIDAL AND PUPICIDAL ACTION OF KITCHEN USED PLANT EXTRACTS AGAINST *Culex* MOSQUITO (DIPTERA: CULICIDAE)

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Current research is conducted to test the efficacy of essential oils extracted from some common plants that are mainly used in kitchen as a larvicidal and pupicidal agent against *Culex* mosquitoes. Oil extraction was done from parts (leaves, branches and rhizome) with Soxhlet's apparatus using petroleum ether as a solvent. The knockdown effect was checked after 12, 24, 36 and 48 hours with different concentrations (50, 100, 150 and 200 ppm) along with a control treatment. The data was analysed by Probit analysis to calculate LC_{50} & LT_{50}. The results showed that the early larval instars (1^{st} & 2^{nd}) were more susceptible than later ones (pupae). Ginger oil was more potent after 12 and 24 h against all life stages (2^{nd}, 3^{rd}, 4^{th} instar larvae & pupae) due to lowest LC_{50} values (171 & 136 ppm, 183 & 174 ppm, 246 & 199 ppm and 273 & 217 ppm for 2^{nd}, 3^{rd}, 4^{th} instar larvae & pupae respectively) followed by peppermint, basil, garlic and neem. However, basil oil released its components after 24 and 48 h and became effective more than others. So, we can use these oils in combination for the better management of mosquitoes.

Keywords: *Culex* mosquitoes, Larvicidal, Pupicidal, Plant oils

INTRODUCTION

Inspite of playing a role as vector of different diseases, mosquitoes also play a role in causing neusance to human beings. As vaccine or treatment is not available for most of the mosquito borne diseases such as dengue and West Nile Virus, so the only solution is to control the vector mosquitoes by synthetic pesticides, plant extracts or source reduction (Ravikumar et al., 2011). Due to increment in awareness regarding the harmful effects of chemicals (Naz et al., 2014), the trend is now gradually shifting towards the plant extracts because these are more environment friendly and less toxic. Many plant extracts as larvicidal, pupicidal or repellent have been studied against the mosquitoes earlier (Ravikumar et al., 2011). Every researcher tried to study their local plants against mosquitoes due to their easy availability (Choochote et al., 2005). So, we also tried to check the efficacy of some plant parts that are commonly used in every house kitchen. Therefore, the mosquitocidal (larvicidal & pupicidal) effect of ginger (*Zingiber officinale*), basil (*Ocimum basilicum*) and peppermint (*Mentha piperita*), neem (*Azadirachta indica*) and garlic (*Allium sativum*) had been studied against *Culex* mosquitoes.

MATERIALS AND METHODS

Mosquito collection: Adult mosquitoes were collected with the help of aspirator from the drainage channels along the roads in Faisalabad, Punjab and carried to the department of Zoology, Government College University, Faisalabad for identification and rearing in the plastic cages.

Rearing of mosquitoes: After identification, the adult mosquitoes were kept in rearing cages for rearing. The male were fed with 10 % glucose solution and females with rat blood (Nasir et al., 2015). After feeding, the females laid eggs and the eggs were hatched out in fresh water at 26±1°C and 75±5% RH inside the lab. After hatching the larvae were fed with fish diet and 2nd, 3^{rd}, 4^{th}instar larvae and pupae were used for the bioassay (Kumar et al., 2011).

Collection of plant materials and oil extraction: Ginger (*Zingiber officinale*), basil (*Ocimum basilicum*) and peppermint (*Mentha piperita*), neem (*Azadirachta indica*) and garlic (*Allium sativum*) were collected from kitchen garden and a shop. This plant material was dried and oils were extracted with Soxhlet's apparatus by using petroleum ether as solvent (Cheng et al., 2009b).

Bioassay process: The extracts were checked for their larvicidal effect against 2^{nd}, 3^{rd}, 4^{th} instar larvae and pupicidal effect against pupae with different concentrations (50, 100, 1500 & 200 ppm) by using twenty larvae/pupae for each treatment. Mortality was checked after different time intervals (12, 24, 36 & 48 hrs) using WHO protocol. The experiments were repeated for three times with a control treatment (Sumroiphon et al., 2006).

Statistical analysis: After correcting the mortality data with Abotts formula, LC_{50} and LT_{50} values were calculated with probit analysis by using Minitab software.

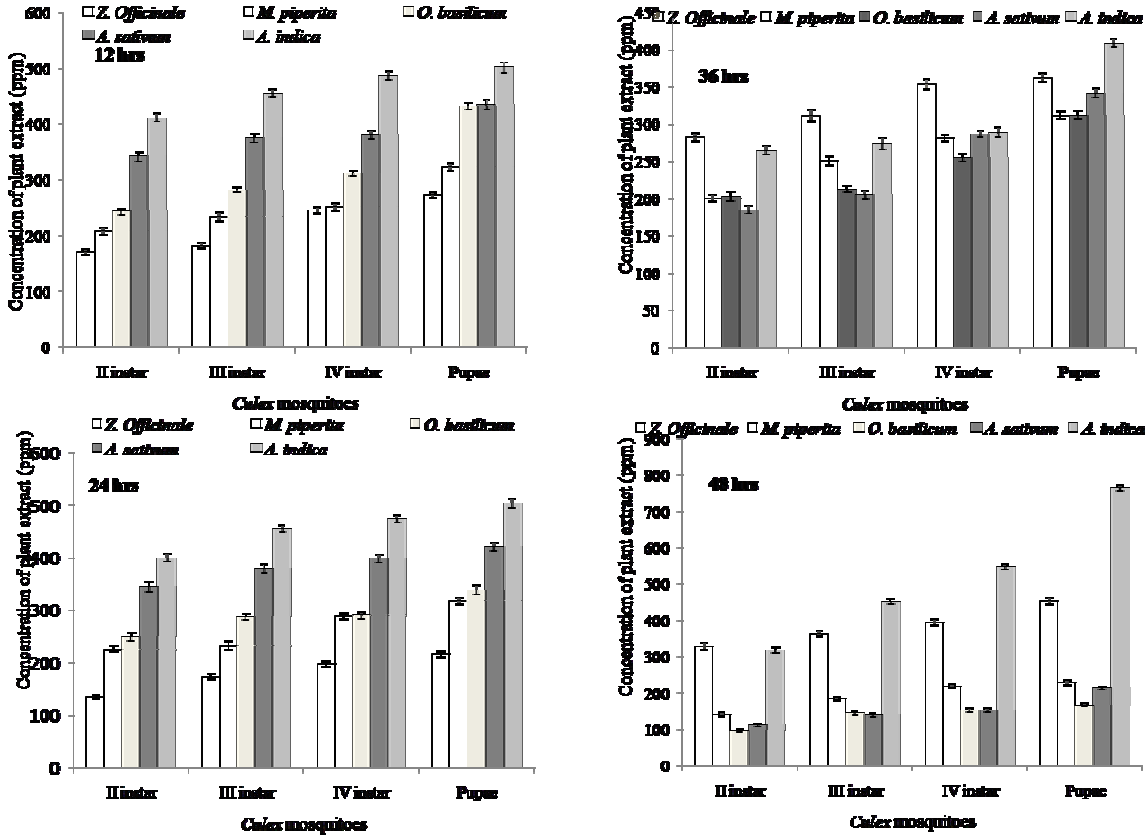

Figure 1. LC_{50} of five plant extracts against different life stages of *Culex* mosquitoes at different time intervals.

RESULTS AND DISCUSSION

Higher mortality was seen in case of 2nd & 3rd instar larvae and least in case of 4th instar larvae and pupae. Slight mortality (5%) was also seen in case of control treatment. In case of oils, the ginger oil showed greatest mortality on the other hand basil showed least mortality in case of all immature stages of *Culex*. The results showed that the potent plant extracts after 12 hrs was *Z. officinale* with least LC_{50} value (171 ppm) for 2nd instar larvae and the least effective plant was *A. indica* with the highest LC_{50} value (412 ppm). In case of 3rd instar larvae, again ginger was more effective with less LC_{50} (183 ppm) and neem was least effective again with 445 ppm LC_{50} value.

The LC_{50} values for all immature stages of this mosquito were comparatively low for basil and higher for neem than other plant extracts as shown in the Fig. 1. The study related LT_{50} showed that the values of LT_{50} were less for early life stages (2nd & 3rd instar) and more time is needed to kill the 50% population of later life stages (4th instar and pupae) as shown in the Table 1. The values of LT_{50} were high for low concentration like 50 ppm and these values decreased as the concentration increases as shown in table 1.

At 100 ppm, the lowest LT_{50} value (39.7 hrs) was for basil while highest value (55.7 hrs) was for peppermint. The LT_{50} values at 150 & 200 ppm for different life stages with different plant extracts were shown in table 2.

The present research indicated that as we increased concentrations (200 ppm), the mortality also increased as compare to lower concentrations (50 & 100 ppm) after higher time intervals like 36 and 48 hrs. These findings are at par with earlier researchers (Jang et al., 2006). The incompletely purified plant oil extracts are less expensive and highly efficacious for the control of mosquitoes rather than the completely purified compounds or extracts (Jang et al., 2002; Tripathi et al., 2002). Essential oils extracted from the medicinal plants may be an alternative source of different life stages of mosquito control agents since they constitute a rich source of bioactive compounds that are biodegradable into nontoxic products and potentially suitable for use in integrated management programmes. In fact, many researchers have reported on the effectiveness of plant essential oils against mosquito larvae and human parasites, and the recent examples are studied by Aivazi and Vijayan (2008) and Nasir et al. (2015). In addition, Ansari et al. (2000) found that application of *M. piperita* oil at 3 ml/m^2 of water surface area resulted in 100% mortality within 24 h for *Culex* mosquito. These results are similar with our results. I also found more than 90 % mortality at 200 ppm after 48 hrs in case of 2nd instar. Some bioactive compounds are present in these plants that have lethal effects for aquatic life stages of mosquito also have biodegradable properties (Kovendan et al., 2008). The solvent used for extraction process also had effects on the efficacy of plant oils against larvae and pupae of mosquitoes

Table 1. Time mortality response of *Culex* larvae and pupae against different plant extracts

Plant extract	Life stages	50 ppm concentration			100 ppm concentration		
		LT_{50}	Slope±SE	P	LT_{50}	Slope±SE	P
Neem (*Azadirechta indica*)	2nd instar	58.5	1.20 ±0.13	0.00	42.7	1.12 ±0.11	0.00
	3rd instar	65	0.75 ±0.14	0.02	52.4	0.82 ±0.20	0.03
	4th instar	76	0.76±0.16	0.05	58.9	0.81±0.15	0.14
	Pupae	85	0.73 ±0.15	0.00	69	1.51±0.15	0.01
Peppermint (*Mentha piperita*)	2nd instar	60.7	1.32 ±0.13	0.00	55.7	1.12 ±0.13	0.00
	3rd instar	66	0.75 ±0.15	0.02	49	0.91 ±0.19	0.03
	4th instar	45	0.73 ±0.17	0.03	49.9	0.81 ±0.15	0.16
	Pupae	89	0.89 ±0.14	0.00	70	1.69 ±0.18	0.01
Basil (*Ocimum basilicum*)	2ndinstar	45.7	1.19 ±0.13	0.00	39.7	1.14 ±0.11	0.00
	3rd instar	57	0.74 ±0.11	0.01	43	0.89 ±0.20	0.05
	4th instar	77	0.72±0.12	0.05	57	0.80±0.15	0.17
	Pupae	83	0.84 ±0.16	0.01	68.2	1.6±0.14	0.01
Ginger (*Zingiber officinale*)	2ndinstar	59	0.73 ±0.13	0.00	47	1.05 ±0.15	0.02
	3rd instar	67	0.75 ±0.13	0.22	53.5	1.03 ±0.21	0.11
	4th instar	74.9	0.99 ±0.15	0.14	64	0.62± 0.20	0.02
	Pupae	78	0.59 ±0.11	0.09	67	0.43 ±0.13	0.21
Garlic	2ndinstar	54	0.72 ±0.13	0.00	41	1.08 ±0.15	0.02
(*Allium sativum*)	3rd instar	60	0.76 ±0.14	0.22	45.5	1.03 ±0.21	0.10
	4th instar	68	0.98 ±0.14	0.14	54	0.65± 0.19	0.02
	Pupae	71,2	0.55 ±0.11	0.09	59	0.43 ±0.11	0.21

Table 2. Time mortality response of *Culex* larvae and pupae against different plant extracts

Plant extract	Life stages	150 ppm concentration			200 ppm concentration		
		LT_{50}	Slope±SE	P	LT_{50}	Slope±SE	P
Neem (*Azadirechta indica*)	2nd instar	38.7	1.12 ±0.12	0.00	22.4	1.15 ±0.12	0.00
	3rd instar	41	0.72 ±0.13	0.01	37	0.73 ±0.13	0.01
	4th instar	49	0.71±0.14	0.14	44	0.73±0.14	0.04
	Pupae	57	0.70 ±0.13	0.11	49	0.72 ±0.13	0.01
Peppermint (*Mentha piperita*)	2nd instar	40.7	1.15 ±0.12	0.00	22.7	1.18 ±0.12	0.00
	3rd instar	45	0.75 ±0.13	0.01	37	0.79 ±0.13	0.01
	4th instar	51	0.70 ±0.15	0.02	44	0.7 ±0.15	0.04
	Pupae	59	0.82 ±0.15	0.01	55	0.86 ±0.15	0.00
Basil (*Ocimum basilicum*)	2ndinstar	27	1.14 ±0.12	0.00	21.7	1.16 ±0.12	0.00
	3rd instar	32	0.71 ±0.13	0.02	35	0.76 ±0.13	0.01
	4th instar	44	0.73±0.15	0.05	67	0.7 1±0.15	0.04
	Pupae	51.5	0.80 ±0.15	0.01	53	0.82 ±0.15	0.01
Ginger (*Zingiber officinale*)	2ndinstar	35.5	0.70 ±0.13	0.00	24.9	0.73 ±0.13	0.00
	3rd instar	43	0.71 ±0.15	0.27	35	0.74 ±0.15	0.25
	4th instar	52	0.92 ±0.16	0.14	38	0.96 ±0.16	0.13
	Pupae	56	0.51 ±0.12	0.08	39	0.58 ±0.12	0.08
Garlic	2ndinstar	30.5	0.71 ±0.13	0.00	25	0.73 ±0.13	0.00
(*Allium sativum*)	3rd instar	34.5	0.72 ±0.15	0.29	34	0.74 ±0.15	0.25
	4th instar	39.4	0.92 ±0.16	0.14	36	0.96 ±0.16	0.13
	Pupae	46	0.51 ±0.12	0.09	40	0.58 ±0.12	0.08

(Anees, 2008). Our work also indicated the same results that the solvent had strong effects for the mortality of different life stages of mosquitoes (5% mortality). This work concludes that the extracts from these kitchen used plants have good mosquitocidal (larvicidal and pupicidal) properties against the *Culex* immature stages under lab conditions. So, it is suggested that these plant extracts should be studied in the field for the control of dengue mosquito.

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